

Store at
-20°C

#14267

SignalSilence® Rheb siRNA I

www.cellsignal.com

Support: 877-678-TECH (8324)
info@cellsignal.comOrders: 877-616-CELL (2355)
orders@cellsignal.comEntrez-Gene ID #6009
UniProt ID #Q15382

New 08/14

For Research Use Only. Not For Use In Diagnostic Procedures.

Species Cross-Reactivity: H

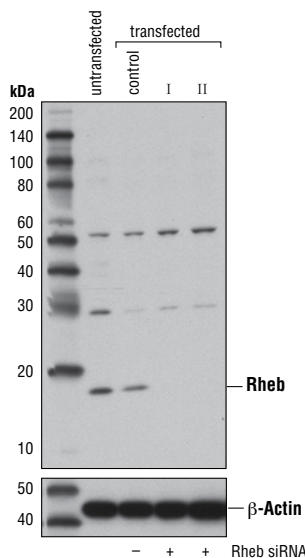
Description: SignalSilence® Rheb siRNA I from Cell Signaling Technology (CST) allows the researcher to specifically inhibit Rheb expression using RNA interference, a method whereby gene expression can be selectively silenced through the delivery of double stranded RNA molecules into the cell. All SignalSilence® siRNA products from CST are rigorously tested in-house and have been shown to reduce target protein expression by western analysis.

Background: Ras Homolog Enriched in Brain (Rheb) is an evolutionarily conserved member of the Ras family of small GTP-binding proteins originally found to be rapidly induced by synaptic activity in the hippocampus following seizure (1). While it is expressed at relatively high levels in the brain, Rheb is widely expressed in other tissues and may be induced by growth factor stimulation. Like other Ras family members, Rheb triggers activation of the Raf-MEK-MAPK pathway (2). Biochemical and genetic studies demonstrate that Rheb has an important role in regulating the insulin/TOR signaling pathway (3-6). The mammalian target of rapamycin (mTOR) is a serine/threonine protein kinase that acts as a sensor for ATP and amino acids, balancing the availability of nutrients with translation and cell growth. The tuberin/hamartin (TSC2/TSC1) complex inhibits mTOR activity indirectly by inhibiting Rheb through the tuberin GAP activity (7).

Directions for Use: CST recommends transfection with 100 nM SignalSilence® Rheb siRNA I 48 to 72 hours prior to cell lysis. For transfection procedure, follow the protocol provided by the transfection reagent manufacturer. Please feel free to contact CST with any questions on use.

Each vial contains the equivalent of 100 transfections, which corresponds to a final siRNA concentration of 100 nM per transfection in a 24-well plate with a total volume of 300 µl per well.

Quality Control: Oligonucleotide synthesis is monitored base by base through trityl analysis to ensure appropriate coupling efficiency. The oligo is subsequently purified by affinity-solid phase extraction. The annealed RNA duplex is further analyzed by mass spectrometry to verify the exact composition of the duplex. Each lot is compared to the previous lot by mass spectrometry to ensure maximum lot-to-lot consistency.



Western blot analysis of extracts from A549 cells, untransfected, transfected with 100 nM SignalSilence® Control siRNA (Unconjugated) #6568 (-), SignalSilence® Rheb siRNA I (+), or SignalSilence® Rheb siRNA II #14284 (+), using Rheb (E1G1R) Rabbit mAb #13879 (upper) and β -Actin (D6A8) Rabbit mAb #8457 (lower). The Rheb (E1G1R) Rabbit mAb confirms silencing of Rheb expression, while the β -Actin (D6A8) Rabbit mAb is used as a loading control.

Storage: Rheb siRNA I is supplied in RNase-free water. Aliquot and store at -20°C.

For product specific protocols and a complete listing of recommended companion products please see the product web page at www.cellsignal.com

Background References:

- (1) Yamagata, K. et al. (1994) *J Biol Chem* 269, 16333-9.
- (2) Yee, W.M. and Worley, P.F. (1997) *Mol Cell Biol* 17, 921-33.
- (3) Inoki, K. et al. (2003) *Genes Dev* 17, 1829-34.
- (4) Stocker, H. et al. (2003) *Nat Cell Biol* 5, 559-65.
- (5) Saucedo, L.J. et al. (2003) *Nat Cell Biol* 5, 566-71.
- (6) Zhang, Y. et al. (2003) *Nat Cell Biol* 5, 578-81.
- (7) Li, Y. et al. (2004) *Trends Biochem Sci* 29, 32-8.

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Applications: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide Species Cross-Reactivity: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—Horse All—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.